

WHAT IS CLAIMED IS:

1. A method for determining whether a compound or agent decreases the activity of a poly(ADP-ribose)-polymerase (PARP) comprising the steps of:

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(a) incubating a mixture comprising:

(i) activated PARP enzyme;

(ii) the compound or agent; and

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(iii) a substrate reagent solution that comprises  $\text{NAD}^+$ ,  $\text{NAD}^+$  having an ADP ribose group labeled with a fluorescence label, DNA, and histone;

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(b) illuminating the mixture of step (a) and a control mixture with plane polarized light having a wavelength at which the fluorescence label fluoresces, and measuring the fluorescence polarization of the mixture of step (a) and the control mixture; and

(c) comparing the measurements of step (b),

wherein the fluorescence polarization measurement of the mixture having a value that is less than the fluorescence polarization measurement of control mixture indicates the compound or agent decreases the activity of the PARP enzyme.

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2. The method of Claim 1, wherein the incubating step (a) has a duration of at least about 10 minutes.

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3. The method of Claim 2, wherein the incubating step has a duration ranging from about 10 minutes to at least about 2 hours.

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4. The method of Claim 1, wherein the fluorescence label comprises phycoerythrin (PE), Texas red (TR), rhodamine, a free lanthanide series salt, a chelated lanthanide series salt, BODIPY, ALEXA, or CyDye.

5. The method of Claim 4, wherein the fluorescence label is Texas red (TR).

6. The method of Claim 5, wherein the wavelength of the plane polarized light is 590 nm.
- 5 7. A method for determining whether a compound or agent decreases the activity of a poly(ADP-ribose)-polymerase (PARP) comprising the steps of:
- (a) incubating a mixture for at least about 10 minutes, wherein the mixture comprises:
- 10 (i) activated PARP enzyme;
- (ii) the compound or agent; and
- (iii) a substrate reagent solution comprising  $\text{NAD}^+$ ,  $\text{NAD}^+$  having an ADP ribose group labeled with a fluorescence label, DNA, and histone;
- 15 (b) illuminating the mixture of step (a) and a control mixture with plane polarized light having a wavelength at which the fluorescence label fluoresces, and measuring the fluorescence polarization of the mixture of step (a) and the control mixture; and
- 20 (c) comparing the measurements of step (b), wherein the fluorescence polarization measurement of the mixture having a value less than the fluorescence polarization measurement of control mixture indicates the compound or agent decreases the activity of the PARP enzyme
- 25 8. The method of Claim 7, wherein the fluorescence label comprises phycoerythrin (PE), Texas red (TR), rhodamine, a free lanthanide series salt, a chelated lanthanide series salt, BODIPY, ALEXA, or CyDye.
- 30 9. The method of Claim 8, wherein the fluorescence label is Texas red, and the wavelength of the plane polarized light is 590 nm.
10. The method of Claim 1, wherein the incubating step has a duration that ranges from about 10 minutes to at least about 2 hours.

11. The method of Claim 9, wherein the  $\text{NAD}^+$  having an ADP ribose group labeled with Texas Red comprises a linker molecule to which the ADP ribose group and the Texas Red are bound.
12. The method of Claim 11, wherein the linker molecule is selected from the group consisting of aminobutyric acid, aminocaproic acid, 7-aminoheptanoic acid, 8-aminocaprylic acid, Fmoc-aminocaproic acid, one or more  $\beta$ -alanines, an isothiocyanate group, a succinimidyl ester, a sulfonal halide, a carbodiimide, and a  $\text{C}_6$  spacer.
13. The method of Claim 12, wherein the linker is the  $\text{C}_6$  spacer.
14. A method for determining whether a compound or agent decreases the activity of a poly(ADP-ribose)-polymerase (PARP) comprising the steps of:
- (a) incubating a mixture that comprises:
    - (i) activated PARP enzyme;
    - (ii) the compound or agent; and
    - (iii) a substrate reagent solution comprising  $\text{NAD}^+$ ,  $\text{NAD}^+$  having an ADP ribose group labeled with Texas Red, DNA, and histone;
  - (b) illuminating the mixture of step (a) and a control mixture with plane polarized light having a wavelength of 590 nm, and measuring the fluorescence polarization of the mixture of step (a) and the control mixture; and
  - (c) comparing the measurements of step (b),
- wherein the fluorescence polarization measurement of the mixture having a value less than the fluorescence polarization measurement of control mixture indicates the compound or agent decreases the activity of the PARP enzyme.
15. The method of Claim 14, wherein the incubating step has a duration of at least about 10 minutes.

16. The method of Claim 15, wherein the incubating step has a duration that ranges from about 10 minutes to at least about 2 hours.
- 5 17. The method of Claim 15, wherein the  $\text{NAD}^+$  having an ADP ribose group labeled with Texas Red comprises a linker molecule to which the ADP ribose group and the Texas Red are bound.
- 10 18. The method of Claim 17, wherein the linker molecule is selected from the group consisting of aminobutyric acid, aminocaproic acid, 7-aminoheptanoic acid, 8-aminocaprylic acid, Fmoc-aminocaproic acid, one or more  $\beta$ -alanines, an isothiocyanate group, an isothiocyanate group, a succinimidyl ester, a sulfonal halide, a carbodiimide, and a  $\text{C}_6$  spacer.
- 15 19. The method of Claim 18, wherein the linker is the  $\text{C}_6$  spacer.
20. The method of Claim 19, wherein the incubating step has a duration of at least 10 minutes.
- 20 21. The method of Claim 20, wherein the step has a duration ranging from about 10 minutes to at least about 2 hours.
- 25 22. A method for determining whether a compound or agent decreases the activity of a poly(ADP-ribose)-polymerase (PARP) comprising the steps of:
- 30 (a) Incubating a mixture for at least 10 minutes, wherein the mixture comprises:
- (i) activated PARP enzyme;
  - (ii) the compound or agent; and
  - (iii) a substrate reagent solution comprising  $\text{NAD}^+$ ,  $\text{NAD}^+$  having an ADP ribose group labeled with Texas Red, DNA, and histone;
- (b) illuminating the mixture of step (a) and a control mixture with plane polarized light having a wavelength of 590 nm, and measuring the fluorescence

polarization of the mixture of step (a) and the control mixture a wavelength of 620 nm; and

(c) comparing the measurements of step (b),

- 5 wherein the fluorescence polarization measurement of the mixture having a value that is less than the fluorescence polarization measurement of control mixture indicates the compound or agent decreases the activity of the PARP enzyme.

10 23. The method of Claim 22, wherein, wherein the  $\text{NAD}^+$  having an ADP ribose group labeled with Texas Red comprises a linker molecule to which the ADP ribose group and the Texas Red are bound.

15 24. The method of Claim 23, wherein the linker molecule is selected from the group consisting of aminobutyric acid, aminocaproic acid, 7-aminoheptanoic acid, 8-aminocaprylic acid, Fmoc-aminocaproic acid, one or more  $\beta$ -alanines, an isothiocyanate group, an isothiocyanate group, a succinimidyl ester, a sulfonal halide, a carbodiimide, and a  $\text{C}_6$  spacer.

20 25. The method of Claim 24, wherein the spacer is the  $\text{C}_6$  spacer.